Declaration of Dr. Harvey Fishman Under 37 CFR § 1.132

1. I, Harvey Fishman, declare as follows, under penalty of perjury.

2. I hold M.S. and Ph.D. degrees in Chemistry and an M.D. degree from Stanford University.

3. I have an extensive research background, including analytical chemistry, design and testing of medical devices, microfabricated structures for biological research, and patch clamp experimentation.

4. I have reviewed US Patent Application No. 2006/0121464 (Jespersen). I have also reviewed US Patent Nos. 7,201,836 (Vogel) and 6,488,829 (Schroeder).

5. I have reviewed the US Patent Office action in Application Serial No. 10/711,327, which contains the statement, referring to Jespersen:

> "although FIG. 3 appears to depict a single aperture, it is interpreted that a plurality of apertures exist on membrane 3 in light of Example 2 which discloses using the device of FIG. 3, [0067], which comprises multiple "sites" which are interpreted to read on the apertures, see [0070-0071]".

This statement is incorrect in light of the patch clamp example given in Jespersen, Example 2. The statement requires two interpretations: 1) a plurality of apertures exist in the membrane and 2) sites and apertures are equivalent. Either interpretation may be individually correct, but they cannot both be correct. The two interpretations are *mutually exclusive*, as follows.

Successful patch clamp measurements have two requirements: an excellent electrical seal across a cell membrane (the "giga-seal") and at least two recording electrodes, one each measuring on both sides of the membrane. The latter requirement is necessary to measure the electrical flow across the membrane, while the former is required to measure the minute changes in current flow.

These two requirements for patch clamp research mean a plurality of recording sites cannot exist without an equivalent plurality of electrodes. One electrode pair is required for each aperture. If there were multiple sites per electrode pair, then: 1) leakage currents would obfuscate the signal, i.e. the combined entity would not likely have a "giga-seal"; and 2) the measurement would not identify which membrane produced a response, and hence there would be no value in studying individual cells instead of the simpler-to-handle ensemble. Note that while each recording site must have at least one electrode, an additional electrode may act as a common electrode across all sites: N aperture electrodes plus 1 common electrode. The minimum number of electrodes is defined as N+1, where N is the number of recording sites.

This reasoning is further evidenced by Vogel and Schroeder. Vogel teaches an example of parallel patch clamp work in which there is more than one aperture. FIG. 7 clearly shows multiple electrodes; in fact, Vogel shows N+1 electrodes, where N is the number of apertures. Similarly, Schroeder teaches a multiple site patch clamp device requiring electrical isolation between recording sites (again, N+1 electrodes are required for simultaneous recording). In FIG. 6, a substrate with an array of apertures is presented. The substrate is joined to a multi-well fixture with matching individual wells. From Schroeder, Col. 8, 44-52:

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> "The purpose of the multi-well fixture is to provide isolated fluidics chambers for each individual hole. The thin substrate is joined to the multi-well fixture…forming an *electrically-isolated* fluid chamber on top of each isolated hole in the substrate."

Jerspersen teaches a single recording electrode on both sides of the membrane, as shown in FIG. 3, equivalent to just 2 electrodes for any number of apertures in the membrane. It is thus not possible in this configuration to do patch clamp recording with multiple apertures; stated differently, if the number of electrodes is N+1=2, then N is necessarily 1. There is only one aperture.

However, an alternate interpretation of Jespersen would be a device to include one electrode per aperture plus one common electrode in an N+1 configuration. This interpretation would require defining the recording "site" in Jespersen as an aperture *plus an electrode*, a very different structure indeed, one which Jespersen does not teach.

Respectfully submitted,

2/11/2010